(a) first and second primers which are specific to and hybridizable with, respectively, first and second nucleic acid sequences which are in opposing strands of human cytomegaloviral DNA (hCMV DNA) and which are separated from each other along said opposing strands by from 90 to 400 nucleotides,

(b) third and fourth primers which are specific to and hybridizable with, respectively, third and fourth nucleic acid sequences which are in opposing strands of a second target DNA which is the same as or different from hCMV DNA, the third and fourth nucleic acid sequences being different from said first and second nucleic acid sequences and being separated from each other along the opposing strands by from 90 to 400 nucleotides,

each of said first, second, third and fourth primers having a T_m within the range of from about 65 to about 74°C, all of said primer T_m's being within about 5°C of each other, said first and second primers having nucleotide lengths which differ from each other by no more than 5 nucleotides, and said third and fourth primers having nucleotide lengths which differ from each other by no more than 5 nucleotides, and each of said first, second, third and fourth primers being present in the same amount within the range of from about 0.1 to about 2 µmolar, and

(c) a thermostable DNA polymerase present at at least 10 units/100 µl.

2. (amended) The composition of claim 1 wherein [each of said primers is present at a concentration of at least about 0.075 µmolar, and] said composition further comprises

[a thermostable DNA polymerase present at from about 0.1 to about 50 units/100 µl,]

a DNA polymerase cofactor present at from about 2 to about 15 mmolar, and

a dNTP present at from about 0.25 to about 3.5 mmolar.

9. (amended) The composition of claim 1 wherein said first (a) and second (b) primers are selected from the group of primer sets consisting of:

Primer set 1:

(a) SEQ ID NO:1 5'-GAGGCTATTG TAGCCTACAC TTTGG-3'

(b) SEQ ID NO:2 5'-CAGCACCATC CTCCTCTTCC TCTGG-3',

and

Primer set 2:

(a) SEQ ID NO:3 5'-TGCACTGCCA GGTGCTTCGG CTCAT-3'

(b) SEQ ID NO:4 5'-CACCACGCAG CGGCCCTTGA TGTTT-3'.

11. (amended) The composition of claim [11] 10 wherein said labeled primers are labeled with biotin.

12. (amended) A diagnostic test kit for the amplification of human cytomegaloviral DNA and a second target DNA comprising, in separate packaging:

a) an aqueous composition buffered to a pH of from about 7 to about 9, which comprises:

first and second primers which are specific to and hybridizable with, respectively, first and second nucleic acid sequences which are in opposing strands of human cytomegaloviral DNA (hCMV DNA) and which are separated from each other along said opposing strands by from 90 to 400 nucleotides,

third and fourth primers which are specific to and hybridizable with, respectively, third and fourth nucleic acid sequences which are in opposing strands of a second target DNA which is the same as or different from hCMV DNA, the third and fourth nucleic acid sequences being different from said first and second nucleic acid sequences and being separated from each other along the opposing strands by from 90 to 400 nucleotides,

each of said first, second, third and fourth primers having a T_m within the range of from about 65 to about 74°C, all of said primer T_m's being within about 5°C of each other, said first and second primers having nucleotide lengths which differ from each other by no more than 5 nucleotides, and said third and fourth primers having nucleotide lengths which differ from each

other by no more than 5 nucleotides, and each of said first, second, third and fourth primers being present in the same amount within the range of from about 0.1 to about 2 μmolar, and

a thermostable DNA polymerase present at at least 10 units/100 μl,

b) at least one additional PCR reagent, and

c) a capture reagent comprising a water-insoluble support to which is covalently attached a capture probe which is specific to a nucleic acid sequence of a strand of hCMV DNA, said capture probe having from 10 to 40 nucleotides and a T_m greater than about 50°C, and is hybridizable with said nucleic acid sequence of said hCMV DNA strand at a temperature in the range of from about 40 to about 55°C, and

a second capture reagent comprising a water-insoluble support to which is covalently attached a second capture probe which is specific to a nucleic acid sequence of a strand of said second target DNA, said second capture probe having from 10 to 40 nucleotides and a T_m greater than about 50°C, and being hybridizable with said nucleic acid sequence of said second target DNA strand at a temperature in the range of from about 40 to about 55°C.

15. (amended) The test kit of claim [14] 12 wherein said first capture probe is selected from the group consisting of:

SEQ ID NO:5 5'-GGTGTCACCC CCAGAGTCCC CTGTACCCGC-3',

SEQ ID NO:6 5'-GACACAGTGT CCTCCCGCTC CTCCTGAGCA-3',

SEQ ID NO:7 5'-GTGGAAGGCG GCTCGCTGGA AGCCGGTCGT-3', and

SEQ ID NO:8 5'-GAACCGAGGG CCGGCTCACC TCTATGTTGG-3'.

16. (amended) The test kit of claim 12 wherein [each of said primers is present at a concentration of at least about 0.075 μmolar, and] said composition further comprises

[a thermostable DNA polymerase present at from about 0.1 to about 50 units/100 μl,]

a DNA polymerase cofactor present at from about 2 to about 15 mmolar, and

a4 a dNTP present at from about 0.25 to about 3.5 mmolar.

19. (amended) A diagnostic test kit for the amplification of human cytomegaloviral DNA and a second target DNA comprising, in separate packaging:

a) an aqueous composition buffered to a pH of from about 7 to about 9, and comprising first and second primers which are specific to and hybridizable with, respectively, first and second nucleic acid sequences which are in opposing strands of hCMV DNA and which are separated from each other along said opposing strands by from 90 to 400 nucleotides,

each of said first and second primers <u>being present in the same amount within the range of from about 0.1 to about 2 µmolar and</u> having a T_m within the range of from about 65 to about 74°C, said primer T_m's being within about 5°C of each other, and

a5 said first and second primers having nucleotide lengths which differ from each other by no more than 5 nucleotides,

b) <u>a separate aqueous composition buffered to a pH of from about 7 to about 9, and comprising</u> third and fourth primers which are specific to and hybridizable with, respectively, third and fourth nucleic acid sequences which are in opposing strands of a second target DNA which is the same as or different from hCMV DNA and which are separated from each other along said opposing strands of said second target DNA by from 90 to 400 nucleotides,

each of said third and fourth primers <u>being present in the same amount of from about 0.1 to about 2 µmolar and</u> having a T_m within the range of from about 65 to about 74°C, said third and fourth primer T_m's being within about 5°C of each other and within about 5°C of the T_m's of said first and second primers, and said third and fourth primers having nucleotide lengths which differ from each other by no more than 5 nucleotides, [and]

c) included in either a) or b), a thermostable DNA polymerase present at at least 10 units/100 µl,

d) at least one additional PCR reagent, and

e) a first capture reagent comprising a water-insoluble support to which is covalently attached a first capture probe which is specific to a nucleic acid sequence of a strand of hCMV DNA, said first capture probe having from 10 to 40 nucleotides and a T_m greater than about 50°C, and is hybridizable with said nucleic acid sequence of said hCMV DNA strand at a temperature in the range of from about 40 to about 55°C, and

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a second capture reagent comprising a water-insoluble support to which is covalently attached a second capture probe which is specific to a nucleic acid sequence of a strand of said second target DNA, said second capture probe having from 10 to 40 nucleotides and a T_m greater than about 50°C, and is hybridizable with said nucleic acid sequence of said second target DNA strand at a temperature in the range of from about 40 to about 55°C,

said first and second capture probes having T_m's which differ by no more than about 15°C.

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22. (amended) A method for the amplification and detection of human cytomegaloviral DNA and a second target DNA comprising:

A) simultaneously subjecting the denatured opposing strands of hCMV DNA and the denatured opposing strands of a second target DNA to polymerase chain reaction in the presence of:

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i) an aqueous composition buffered to a pH of from about 7 to about 9, and comprising

first and second primers which are specific to and hybridizable with, respectively, first and second nucleic acid sequences which are in opposing strands of hCMV DNA and which are separated from each other along said opposing strands by from 90 to 400 nucleotides,

third and fourth primers which are specific to and hybridizable with, respectively, third and fourth

nucleic acid sequences which are in opposing strands of a second target DNA which is the same as or different from hCMV DNA, the third and fourth nucleic acid sequences being different from said first and second nucleic acid sequences and being separated from each other along the opposing strands by from 90 to 400 nucleotides,

each of said first, second, third and fourth primers having a T_m within the range of from about 65 to about 74°C, all of said primer T_m's being within about 5°C of each other, said first and second primers having nucleotide lengths which differ from each other by no more than 5 nucleotides, and said third and fourth primers having nucleotide lengths which differ from each other by no more than 5 nucleotides, and each of said first, second, third and fourth primers being present in the same amount within the range of from about 0.1 to about 2 μmolar, and

ii) the following additional PCR reagents: a thermostable DNA polymerase present in an amount of at least 10 units/100 μl, a DNA polymerase cofactor and at least one dNTP, any or all of said additional PCR reagents being in the same or a different composition as defined in i),

to simultaneously amplify said opposing hCMV DNA strands and the opposing second target DNA strands wherein, in each PCR cycle, priming and primer extension are carried out at the same temperature within the range of from about 62 to about 75°C and carried out within 120 seconds, [and]

B) capturing one of said amplified hCMV DNA strands with a capture reagent comprising a water-insoluble support to which is covalently attached a capture probe which is specific to a nucleic acid sequence of said hCMV DNA strand, said capture probe having from 10 to 40 nucleotides and a T_m greater than about 50°C, and is hybridizable with said nucleic acid sequence of said hCMV DNA strand at a temperature in the range of from about 40 to about 55°C, and

capturing one of said amplified second target DNA strands with a second capture reagent comprising a second capture probe specific to a nucleic acid sequence of said second target DNA strand, said second capture probe having from 10 to 40 nucleotides and a T_m greater than about 50°C, and is hybridizable with said nucleic acid sequence of said second target DNA strand at a temperature in the range of from about 40 to about 55°C,

said first and second capture probes having T_m's which differ by no more than about 15°C, and

C) simultaneously detecting [at least one of] said captured amplified hCMV DNA [strands] strand and [at least one of the] said captured amplified second target DNA strands as a simultaneous determination of the presence of hCMV DNA and [the] said second target DNA.

27. (amended) The method of claim [23] 22 wherein PCR is carried out for from 20 to 50 cycles.

30. (amended) The method of claim [29] 22 wherein said water-insoluble support for each capture reagent is a polymeric or magnetic particle having a diameter in the range of from about 0.001 to 10 μmeters, and each of said capture probes has a T_m greater than about 55°C.

31. (amended) The method of claim [29] 22 wherein said first and second capture reagents are disposed in distinct regions on a water-insoluble substrate of a test device.

32. (amended) The method of claim [29] 22 wherein said first capture probe is selected from the group consisting of:

SEQ ID NO:5 5'-GGTGTCACCC CCAGAGTCCC CTGTACCCGC-3',

SEQ ID NO:6 5'-GACACAGTGT CCTCCCGCTC CTCCTGAGCA-3',

SEQ ID NO:7 5'-GTGGAAGGCG GCTCGCTGGA AGCCGGTCGT-3', and

SEQ ID NO:8 5'-GAACCGAGGG CCGGCTCACC TCTATGTTGG-3'.

33. (amended) The method of claim [23] 22 wherein said second target DNA is selected from the group consisting of a retroviral DNA, *Mycobacterium tuberculosis* DNA, *Mycobacterium avium* DNA, Epstein Barr

viral DNA, respiratory syncytial viral DNA, *Pneumocystis carinii* DNA and hepatitis DNA.

34. (amended) The method of claim [23] 22 wherein said first (a) and second (b) primers are selected from the group of primer sets consisting of:

Primer set 1:

(a) SEQ ID NO:1 5'-GAGGCTATTG TAGCCTACAC TTTGG-3'

(b) SEQ ID NO:2 5'-CAGCACCATC CTCCTCTTCC TCTGG-3',

and

Primer set 2:

(a) SEQ ID NO:3 5'-TGCACTGCCA GGTGCTTCGG CTCAT-3'

(b) SEQ ID NO:4 5'-CACCACGCAG CGGCCCTTGA TGTTT-3',

and

said first capture probe is selected from the group consisting of:

SEQ ID NO:5 5'-GGTGTCACCC CCAGAGTCCC CTGTACCCGC-3',

SEQ ID NO:6 5'-GACACAGTGT CCTCCCGCTC CTCCTGAGCA-3',

SEQ ID NO:7 5'-GTGGAAGGCG GCTCGCTGGA AGCCGGTCGT-3', and

SEQ ID NO:8 5'-GAACCGAGGG CCGGCTCACC TCTATGTTGG-3'.

35. (amended) The method of claim [23] 22 wherein [each of said primers is present at a concentration of at least about 0.075 μmolar,

a thermostable DNA polymerase is present at from about 0.1 to about 50 units/100 μl,]

a DNA polymerase cofactor is present at from about 2 to about 15 mmolar, and

a dNTP is present at from about 0.25 to about 3.5 mmolar.

36. (amended) The method of claim [23] 22 wherein a third target DNA is amplified and detected simultaneously with the hCMV DNA and said second target DNA,

said third target DNA being amplified using a third set of primers wherein each primer has a T_m within the range of from about 65 to about 74°C, the primer T_m's being within 5°C of each other and within 5°C of said first, second, third and fourth primers, and the lengths of primers in said third primer set differing by no more than 5 nucleotides.

38. (amended) An oligonucleotide [defined as] having the sequence:

SEQ ID NO:3 TGCACTGCCA GGTGCTTCGG CTCAT,
SEQ ID NO:16 CATTCCCACT GACTTTCTGA CGCACGT,
SEQ ID NO:17 TGAGGTCGTG GAACTTGATG GCGT,
SEQ ID NO:18 GGTCATCGCC GTAGTAGATG CGTAAGGCCT,
SEQ ID NO:19 GGAATGACGC AAGGACATAT GGGCGT,
SEQ ID NO:20 CCCAGGTGCA CACCAATGTG GTGGAT,
SEQ ID NO:21 GGACTGTGCG CGTTGTATAC CCTGC,
SEQ ID NO:22 ACTCCCGAAG CGAATGGCAC GTGGA,
SEQ ID NO:23 CATAGCTTGT GCCCGTGTGG CACGT,
SEQ ID NO:24 CCAAGACGAG ACCGTCAGAG CTGGT,
SEQ ID NO:25 AAGCTGTTGC CGCCATCAAA TAAACG, or
SEQ ID NO:26 CTGCGTTAGA CCGAGAACTG TGGATAAAGG.

REMARKS

The specification has been amended on pages 33 and 45 to correct two typographical errors. In addition, the status of the allowed application mentioned on page 4 has been updated.

The title of the application has not been amended at this time. There is no requirement in the MPEP that the title be 17 words or less. It suggests as short a title as possible. Applicants believe the current title is properly descriptive of the claimed invention. If the Examiner wants to suggest a shorter title which is equally descriptive, Applicants are willing to consider it.

Likewise, there is no requirement that Figures 1-8 contain description of the X and Y axes on the drawings themselves. Applicants have provided the appropriate descriptions in the Examples. Specifically, Figures 1-6 are described in the paragraph bridging pages 37-38, and Figures 7 and 8 are described in the last paragraph on page 38. This is sufficient. Applicants have declined to put English text on US application figures where it is not needed so the same figures can be used in foreign applications without need for translation of the English descriptions.